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IMMUNOHISTOCHEMICAL REACTIVITY OF ANTI-LAV p18 MONOCLONAL ANTIBODY IN LYMPHNODES FROM PGL AND AIDS PATIENTS

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Summary - This study deals with the immunohistochemistry of a monoclonal antibody (Mab) raised against the p18 protein of the LAV virus in lymphnodes from 20 cases of persistent generalized lymphoadenopathy (PGL) (2) and 6 of acquired immunodeficiency syndrome (AIDS) (3).

In all the PGL cases that have been studied, we observed a very important disruption of the follicular dendritic cell's (FDC) framework in the germinal centers which is associated with a strong positivity of anti-LAV p18 Mab. This finding was always associated with the presence of an increased number of Leu2a+ lymphocytes inside the germinal centers. This observation is consistent with the main morphological feature of the lymphnodes in the PGL syndrome which is mainly characterized by lesions of FD cells.

Riassunto - Abbiamo studiato la reattività immunoistochimica di un monoclonale diretto contro la proteina p18 del virus LAV in linfonodi di pazienti con PGL o AIDS. In tutti i casi studiati la positività era limitata solo alle cellule dendritiche follicolari la cui trama reticolare era sovvertita o distrutta, come d'altronde è da attendersi in questo tipo di patologia.

Questo reperto si associa alla presenza di numerosi linfociti con fenotipo Leu2a+ all'interno dei centri germinativi.

Introduction

Lymphnode pathology in HTLVIII/LAV virus infection is characterized by a generalized lymphnodal enlargement, with the histological feature of a very important follicular hyperplasia (4,6); in AIDS patients, on the contrary, only a severe depletion of lymphoid cells and no follicular reactivity is observable (12).

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Viral antigens have been detected in lymphoid cells bearing OKT4 immunophenotype by many Authors (5). We have tested a LAV specific monoclonal antibody (Mab) which recognizes an epitope present on the precursor GAG protein of 55 KD and on some of its cleavage products, namely p18 and p40 (G. Chassagne *et al*, manuscript submitted on frozen sections of lymphnodes from 20 cases of PGL and 6 cases of AIDS.

Materials and Methods

Lymph nodal biopsies from 20 PGL and from 6 AIDS patients have been studied. In all the cases the clinical diagnosis was established according to the CDC criteria (2,3). In PGL cases, the histological diagnosis was of follicular hyperplasia with variable degrees of paracortical reactivity. A severe lymphoid depletion with hyaline involution, or total fading out was evident in all but one AIDS cases, where a few secondary follicles were still present.

Five non tumoral lymphnodes with follicular hyperplasia served as controls.

Immunohistochemical procedures were performed on frozen sections using a triple step ABC technique (11).

The anti-LAV p18 monoclonal antibody was obtained at the Pasteur Institute (Paris, France).

Several commercially available monoclonal antibodies were also employed: DRC-1, Pan-B (Dako immunoglobulin, Copenhagen, Denmark); Leu 2a, Leu 3a (Becton Dickinson Laboratory Systems, Mechelen, Belgium). Each Mab was simultaneously tested on lymphnodal sections from the control group and the PGL and AIDS patients collected on the same slide in order to avoid misinterpretations due to technical pitfalls or unexpected cross reactions.

Results

Immunohistochemical reactivity of PGL lymphnodes was characterized by the disruption of the follicular dendritic cell (FDC) framework as reported by Janossi *et al.* (1985), well emphatised by the Mab DRC-1, and by the presence of many lymphocytes

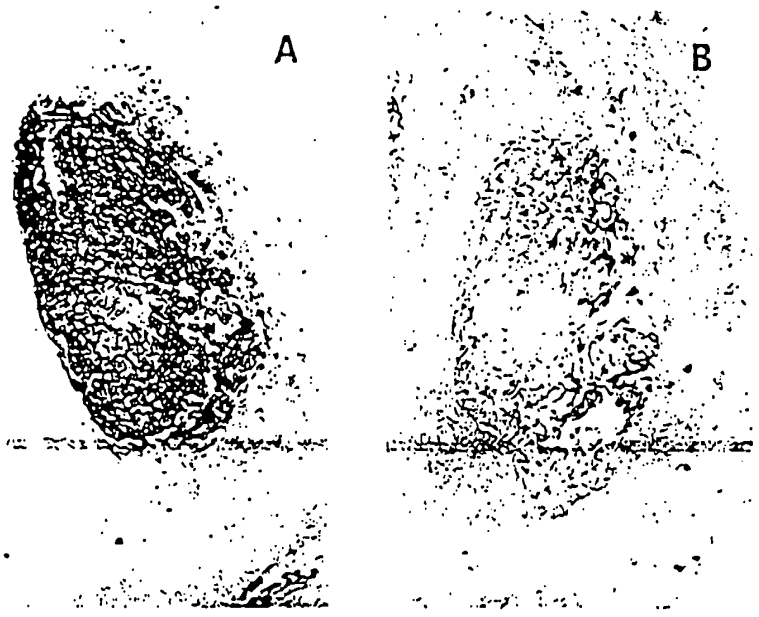

Figure 1 - Lymphnode from PGL patient. Serial frozen sections stained with DRC-1 (A) and anti-LAV p18 (B) monoclonal antibodies.

The staining pattern obtained with the anti-LAV p18 Mab is not different from that obtained with the DRC-1 Mab specific for FD cells. Note in (B) a stronger staining of the disrupted areas. ABC immunoperoxidase method. No counterstain (250X).

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bearing Leu 2a+ immuno-phenotype inside the follicular germinal centers.

In all the PGL cases the positivity of Mab anti-LAV p18 was restricted to the follicular germinal centers, with a staining pattern indicating that the involvement was limited to the FDC.

A stronger positivity was observed in those follicles where the disruption of the FDC framework was more evident.

No other lymphnodal cell, neither lymphoid nor vascular, was evidenced.

No reactivity was observed with Mab DRC-1 and anti-LAV p18 in five of the AIDS lymphnodes characterized by hyaline involution or total loss of the germinal centres.

In one case, however, where a follicular reactivity was still present, the FDC framework was positive with both DRC-1 and anti-LAV p18 Mabs.

In the control group no disruption of the FDC framework was evidenced with the DRC-1 Mab, and lymphocytes bearig Leu2a immuno-phenotype inside the germinal centers were rare or absent. Mab anti-LAV p18 was unreactive in all the cases we have tested.

Discussion

Ultrastructural evidence of retrovirus-like particles in lymphnodes from AIDS and PGL patients has been reported by some Authors (1, 13).

Our results with the Mab anti-LAV p18 support these observations. Immunohistochemical staining on lymphnodal frozen sections, however, shows a generalized involvement of the FDC, without positivity in any other cell.

The presence of the LAV p18 antigen in, or on, the surface of FDC could be interpreted in several ways. The fact that the main morphological aspect of lymphnodes in HTLV III/LAV virus infection is due to the loss of FDC framework, suggests a correlation between these phenomena. Furthermore, the presence of an increased number of Leu2a+ lymphocytes inside the germinal centers (8, 9, 10, 11), must have a significance, although it is, until now, still obscure. One could reasonably hypothesize that the presence of Leu2a+ cytotoxic cells has something to do with the death of FDC, perhaps through an immune mechanism elicited by the presence of the viral antigens presented by them.

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